

SignalSilence® Puma siRNA I

✓ 10 µM in 300 µl
(3 nmol)

Cell Signaling
TECHNOLOGY®

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For Research Use Only. Not For Use In Diagnostic Procedures.

Species Cross-Reactivity: H, (Mk)

Description: SignalSilence® Puma siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit Puma expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence® siRNA products from CST are rigorously tested in-house and have been shown to reduce target protein expression by western analysis.

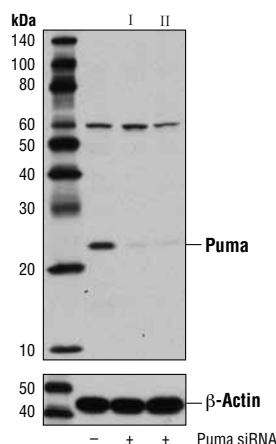
Background: Puma (p53 upregulated modulator of apoptosis) is a "BH3-only" Bcl-2 family member originally identified in differential gene expression studies as a p53-inducible gene (1,2). The "BH3-only" family members include Bad, Bid, Bik, Bcl-2, Bim, and Noxa, all of which contain a BH3 domain but lack other conserved domains, BH1 and BH2, and generally promote apoptosis by binding to and antagonizing anti-apoptotic Bcl-2 family members through BH3 domain interactions (3). Two BH3-containing proteins are produced from the puma gene, Puma-α and Puma-β, both of which are induced by p53, bind Bcl-2 and Bcl-xL, localize to the mitochondria, and promote cytochrome c release and apoptosis (1,2). Puma plays a critical role in the p53 tumor suppressor pathway. Targeted disruption of the puma gene impairs p53-mediated apoptosis and tumor suppression (4-7). Puma knockout mice show defects from multiple apoptotic stimuli, including ionizing irradiation, deregulated c-Myc expression, and cytokine withdrawal (4).

Specificity/Sensitivity: SignalSilence® Puma siRNA I inhibits human and monkey Puma expression.

Directions for Use: CST recommends transfection with 100 nM SignalSilence® Puma siRNA I 48 to 72 hours prior to cell lysis. For transfection procedure, follow protocol provided by the transfection reagent manufacturer. Please feel free to contact CST with any questions on use.

Each vial contains the equivalent of 100 transfections, which corresponds to a final siRNA concentration of 100 nM per transfection in a 24-well plate with a total volume of 300 µl per well.

Quality Control: Oligonucleotide synthesis is monitored base by base through trityl analysis to ensure appropriate coupling efficiency. The oligo is subsequently purified by affinity-solid phase extraction. The annealed RNA duplex is further analyzed by mass spectrometry to verify the exact composition of the duplex. Each lot is compared to the previous lot by mass spectrometry to ensure maximum lot-to-lot consistency.



Western blot analysis of extracts from A549 cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Puma siRNA I (+), or SignalSilence® Puma siRNA II #12890 (+), using Puma (D30C10) Rabbit mAb #12450 (upper) or β-Actin (D6A8) Rabbit mAb #8457 (lower). The Puma (D30C10) Rabbit mAb confirms silencing of Puma expression, while the β-Actin (D6A8) Rabbit mAb is used as a loading control.

Entrez-Gene ID #27113

UniProt ID #Q9BXH1

Storage: Puma siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

Please visit www.cellsignaling.com for a complete listing of recommended companion products.

Background References:

- (1) Yu, J. et al. (2001) *Mol. Cell* 7, 673-682.
- (2) Nakano, K. and Vousden, K.H. (2001) *Mol. Cell* 7, 683-694.
- (3) Bouillet, P. and Strasser, A. (2002) *J. Cell Sci.* 115, 1567-1574.
- (4) Jeffers, J.R. et al. (2003) *Cancer Cell* 4, 321-328.
- (5) Hemann, M.T. et al. (2004) *Proc. Natl. Acad. Sci. USA* 101, 9333-9338.
- (6) Yu, J. et al. (2003) *Proc. Natl. Acad. Sci. USA* 100, 1931-1936.
- (7) Villunger, A. et al. (2003) *Science* 302, 1036-1038.

Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide

Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine

Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.